

WE CLAIM:

1. A method of determining the optimal level of product expression in animal cell culture wherein the concentration of a solute of interest in a culture medium composition for optimal product expression is different than the culture medium composition determined for optimal cell growth, which method comprises:

- a) growing the animal cell culture in medium to determine optimal cell growth;
- b) varying the concentration of the solute in the culture medium to a concentration above that optimal for cell growth which concentration is effective to create an environment of solute stress on the cell culture;
- c) monitoring the product expression under the varying solute concentration conditions to determine optimal product expression; and
- d) selecting the solute concentration that provides the optimal combination of cell growth and product expression which allows for optimal productivity.

2. The method of claim 1, wherein the concentration of said solute that provides the optimal combination of cell growth and product expression causes a decrease in cell growth rate or maximum cell density.

3. The method of claim 2, wherein said animal cell culture is a mammalian cell culture.

4. The method of claim 3, wherein said mammalian cell culture is a hybridoma cell culture that expresses monoclonal antibodies.

5. The method of claim 4, wherein the hybridoma cell culture produces IgM or IgG monoclonal antibodies.

6. The method of claim 3, wherein said monoclonal antibodies are human or murine monoclonal antibodies.

7. The method of claim 5, wherein the hybridoma is selected from the group consisting of D-234 (ATCC HB-8598), D-234 (ATCC HB-9543), T-88 (ATCC HB-9431), and 454A12 (IVI 10075).

8. The method of claim 2, wherein said solute is an inorganic salt or ion thereof.

9. The method of claim 8, wherein said inorganic salt or ion thereof includes sodium chloride and potassium chloride.

10. The method of claim 9, wherein the mammalian cell culture is composed of D-234 cells and the osmolality of the medium with the addition of sodium chloride is in the range of 350 to 400 mOsmol/kg.

11. The method of claim 9, wherein the mammalian cell culture is composed of T-88 cells and the osmolality of the medium with the addition of sodium chloride is in the range of 400 to 450 mOsmol/kg.

12. The method of claim 2, wherein said solute is a metabolite.

13. The method of claim 12, wherein said metabolite includes lactate and ammonium.

14. The method of claim 13, wherein lactate is added as sodium lactate and the sodium lactate concentration is in the range of 10 to 100 mM.

15. The method of claim 14, wherein the sodium lactate concentration is in the range of 40 to 60 mM.

16. The method of claim 15, wherein said animal cell culture is a mammalian hybridoma cell culture that expresses monoclonal antibodies.

17. The method of claim 16, wherein said mammalian hybridoma cell culture is selected from the group consisting of D-234 (ATCC HB-8598), D-234 (ATCC HB-9543), T-88 (ATCC HB-9431), and 454A12 (IVI 10075).

18. The method of claim 13, wherein ammonium is added as ammonium chloride and the ammonium chloride concentration is in the range of 3 to 20 mM.

19. The method of claim 18, wherein the ammonium chloride concentration is in the range of 10-15 mM.

20. The method of claim 19, wherein said animal cell culture is a mammalian hybridoma cell culture that expresses monoclonal antibodies.

21. The method of claim 20, wherein said mammalian hybridoma cell culture is selected from the group consisting of D-234 (ATCC HB-8598), D-234 (ATCC HB-9543), T-88 (ATCC HB-9431), and 454A12 (IVI 10075).

22. The method of claim 2, wherein the solute is an organic polyol.

23. The method of claim 22, wherein the organic polyol is glucose.

24. The method of claim 23, wherein the glucose concentration is in the range of 7-15 g/l.

25. The method of claim 24, wherein said animal cell culture is a mammalian hybridoma cell culture that expresses monoclonal antibodies.

26. The method of claim 25, wherein said mammalian hybridoma cell culture is selected from the group consisting of D-234 (ATCC HB-8598), D-234 (ATCC HB-9543), T-88 (ATCC HB-9431), and 454A12 (IVI 10075).

27. A method of increasing the production of monoclonal antibodies during mammalian cell culture comprising culturing hybridoma cells under conditions of solute stress.

28. The method of claim 27, wherein solute stress is produced by an increased concentration of a solute selected from the group consisting of inorganic salts or ions thereof, metabolites, and organic polyols.

29. The method of claim 28, wherein the monoclonal antibodies are selected from the group consisting of: IgM monoclonal antibodies and IgG monoclonal antibodies.

30. The method of claim 29, wherein said mammalian hybridoma cell culture is selected from the group consisting of D-234 (ATCC HB-8598), D-234 (ATCC HB-9543), T-88 (ATCC HB-9431), and 454A12 (IVI 10075).

31. The method of claim 28, wherein the concentration of said solute causes a decrease in cell growth.

32. The method of claim 22, wherein the organic polyol is polypropylene glycol.

34. The method of claim 32, wherein the polypropylene glycol concentration is about 8 $\mu\text{l/L}$.

36. The method of claim 34, wherein said mammalian hybridoma cell culture is selected from the group consisting of D-284 (ATCC HB-8598), D-234 (ATCC HB-9543), T-88 (ATCC HB-9431), and 454A12 (IVI 10075).

38. The method of claim 9, wherein the mammalian cell culture is comprised of 454A12 hybridomas, and the osmolality of the medium with the addition of sodium chloride is about 400 mOsmol/kg.

Introduction